**510(k) SUBSTANTIAL EQUIVALENCE DETERMINATION**
**DECISION SUMMARY**
**ASSAY ONLY TEMPLATE**

**A. 510(k) Number:**

k051969

**B. Purpose for Submission:**

New Device

**C. Measurand:**

Adrenaline (epinephrine), Noradrenaline (norepinephrine) and dopamine

**D. Type of Test:**

Quantitative Radio Immunoassay (RIA)

**E. Applicant:**

IBL Immuno Biological Laboratories

**F. Proprietary and Established Names:**

TriCat RIA (Adrenaline/ Noradrenaline/Dopamine)

KatCombi RIA (Adrenaline/ Noradrenaline)

Adrenaline RIA

Noradrenaline RIA

**G. Regulatory Information:**

1. Regulation section:

   21 CFR §862.1165, Catecholamines (total) test system

2. Classification:

   Class I, meets the limitation to the exemption, 21 CFR 862.9 (c), (1)

3. Product code:

CHQ, chromatographic/fluorometric method, catecholamines

4. Panel:

   75 Clinical Chemistry

## H. Intended Use:

1. Intended use(s):

   See Indications for use below.

2. Indication(s) for use:

   The IBL Catecholamine RIA test kits are for the in-vitro-diagnostic quantitative determination of the catecholamines adrenaline (epinephrine), noradrenaline (norepinephrine) and/or dopamine (either separate or combined) in human plasma and urine.

   The Catecholamine test kits are useful as an aid in the diagnosis as well as follow-up of tumor diseases of the nervous system, especially of the pheochromocytoma, but also the neuroblastoma and the ganglioneuroma.

3. Special conditions for use statement(s):

   For Prescription Use only

4. Special instrument requirements:

   Gamma Counter

## I. Device Description:

The IBL Catecholamine RIA test kits consist of the following reagents/biological sources for 50 plasma and/or urine tests: lyophilized $^{125}$I-Tracer for adrenaline, noradrenaline and dopamine; lyophilized antiserum (rabbit) for adrenalinee, noradrenaline and dopamine; Precipitating Antiserum (goat); acylation tubes for adrenalinee, noradrenaline and dopamine; extraction plate (macrotiter plate coated with boronate affinity gel); extraction buffer; HCL (0.05 M); lyophilized COMT (catechol-O-methyltransferase ,porcine liver); Coenzyme solution (S-Adenosyl-L-Methionine); Enzyme buffer (Tris buffer, HCL); Standard buffer (1.0 M NaON); Dopamine S/U-Additive (0.125 M HCL); Standards A-F; Controls 1and 2; and Adhesive foil.

## J. Substantial Equivalence Information:

1. Predicate device name(s):

   BioRad Catecholamines By HPLC

   Bioanalytical systems Free Catecholamines & Metanephrines (HPLC)

2. Predicate 510(k) number(s):

   Plasma: k894966 (BioRad); k972167 (Bioanalytical Systems)

   Urine: k873371 (BioRad); k943099 (Bioanalytical Systems)

3. Comparison with predicate:

| **Similarities** | | |
|---|---|---|
| Item | Device | Predicate |
| Analyte | Adrenaline, noradrenaline and dopamine | Adrenaline, noradrenaline and dopamine |
| Matrix | Plasma and Urine | Plasma and Urine |
| Indications for use | Diagnosis/follow-up of tumor diseases of the nervous system, e.g. pheochromocytoma, neuroblastoma and ganglioneuroma | Diagnosis/follow-up of tumor diseases of the nervous system, e.g. pheochromocytoma, neuroblastoma and ganglioneuroma |
| Calculation | Quantitative determination with standard curve | Quantitative determination with standard curve |

| **Differences** | | |
|---|---|---|
| Item | Device | Predicate |
| Method | RIA | HPLC |
| Sample Preparation | Extraction with boronate affinity gel and acylation | Extraction with aluminum oxide |
| Detection | $^{125}$I (Gamma Counter) | Electrochemical detector |

## K. Standard/Guidance Document Referenced (if applicable):

None Referenced

## L. Test Principle:

The devices are complete kits for the measurement of the catecholamines adrenaline, noradrenaline and dopamine by Radio Immunoassay (RIA) based on a competition principle. The specific antibody is bound on the wells of the macrotiter plate. Samples, standards and controls along with extraction buffer are pipetted into the wells on the plate and incubated at room temperature for 30 minutes. The sample wells are emptied and washed. Next, HCl is added to each well and incubated for 15 minutes. Samples, tracer and antiserum are pipetted into acylation tubes for adrenaline, noradrenaline or dopamine and incubated overnight. Antigen in the sample competes with $^{125}$I-labelled antigen. After separation of the bound from the free $^{125}$I-labelled antigen by precipitation and centrifugation, the bound radioactivity is measured in a Gamma counter. Results are determined directly using a standard curve.

## M. Performance Characteristics (if/when applicable):

1. Analytical performance:

   *a. Precision/Reproducibility:*

   Intra assay precision was assessed using urine and plasma control samples. Intra-assay precision was assessed by running Noradrenaline in 2 runs/2 operators one day, Adrenaline in 3 runs/2 operators over 2 days and Dopamine in 2 runs/2 operators one day. Inter-assay precision was assessed by assaying urine and plasma control samples for Noradrenaline in 38 runs/7 operators using 38 lot numbers over 36 days, Adrenaline in 39 runs/5 operators using 37 lot numbers over 37 days and Dopamine in 35 runs/ 7 operators using 35 lot numbers over 33 days. The results are presented in the tables below:

   Intra-assay Precision

| | | Urine 3 | Urine 4 | Plasma 3 | Plasma 4 |
|---|---|---|---|---|---|
| Noradrenaline (ng/mL) | n | 16 | 12 | 16 | 16 |
| | Mean | 54.5 | 126 | 0.324 | 0.951 |
| | SD | 3.0 | 13.7 | 0.020 | 0.082 |
| | %CV | 5.5 | 10.9 | 6.3 | 8.6 |
| Adrenaline (ng/mL) | n | 16 | 18 | 7 | 7 |
| | Mean | 27.4 | 8.8 | 0.350 | 0.977 |
| | SD | 1.8 | 0.7 | 0.016 | 0.056 |
| | %CV | 7.8 | 7.8 | 4.6 | 5.8 |
| Dopamine (ng/mL) | n | 16 | 7 | 16 | 16 |
| | Mean | 364 | 757 | 0.502 | 1.688 |
| | SD | 25 | 104 | 0.043 | 0.180 |
| | %CV | 6.9 | 13.7 | 8.6 | 10.7 |

Inter-assay Precision

| | | Urine | Urine | Urine | Plasma | Plasma | Plasma |
|---|---|---|---|---|---|---|---|
| Noradrenaline (ng/mL) | Days | 36 | 36 | 36 | 36 | 36 | 36 |
| | n | 20 | 20 | 20 | 19 | 19 | 19 |
| | Mean | 31.9 | 141.5 | 287.7 | 0.664 | 1.331 | 3.631 |
| | SD | 3.6 | 15.1 | 35.5 | 0.057 | 0.172 | 0.423 |
| | %CV | 11.2 | 10.7 | 12.3 | 8.6 | 12.9 | 11.6 |
| Adrenaline (ng/mL) | Days | 37 | 37 | 37 | 37 | 37 | 37 |
| | n | 20 | 20 | 20 | 20 | 18 | 19 |
| | Mean | 3.8 | 10.2 | 98.5 | 0.213 | 0.461 | 1.586 |
| | SD | 0.7 | 1.4 | 15.7 | 0.029 | 0.061 | 0.203 |
| | %CV | 18.7 | 13.2 | 15.9 | 13.6 | 13.2 | 12.8 |
| Dopamine (ng/mL) | Days | 33 | 33 | 33 | 33 | 33 | 33 |
| | n | 19 | 20 | 20 | 17 | 11 | 17 |
| | Mean | 79.5 | 365.2 | 747.1 | 0.506 | 2.391 | 4.949 |
| | SD | 15.7 | 33.3 | 91.3 | 0.097 | 0.389 | 0.863 |
| | %CV | 19.8 | 9.1 | 12.2 | 19.2 | 16.2 | 17.4 |

*b. Linearity/assay reportable range:*

The linearity was assessed by assaying serial dilutions of urine and plasma samples having different concentrations for each analyte. Urine samples are automatically diluted during the testing procedure. After the result is read off the standard curve, the urine result is multiplied by 40 for Noradrenaline and Adrenaline, 240 for Dopamine. Urine results below represent values after the calculation. The linearity claim is based on recovery percent deviation of ±25% in linearity across the range. The results obtained were as follows:

| **Noradrenaline** ng/mL | Dilution | 1/1 | 1/2 | 1/4 | 1/8 | 1/16 |
|---|---|---|---|---|---|---|
| Sample | | | | | | |
| Urine 1 | Measured | 118.6 | 62.2 | 34.4 | 17.8 | 8.3 |
| | Expected | | 59.3 | 29.7 | 14.8 | 7.4 |
| | % Recovery | | 105 | 116 | 120 | 112 |
| Urine 2 | Measured | 52.1 | 26.8 | 13.8 | 7.2 | 2.9 |
| | Expected | | 26.1 | 13.0 | 6.5 | 3.3 |
| | % Recovery | | 103 | 106 | 111 | 88 |
| Urine 3 | Measured | 69.4 | 34.4 | 19.1 | 9.5 | 5.3 |
| | Expected | | 34.7 | 17.4 | 8.7 | 4.3 |
| | % Recovery | | 99 | 110 | 109 | 123 |

| *Noradrenaline* ng/mL | Dilution | 1/1 | 1/2 | 1/4 | 1/8 | 1/16 |
|---|---|---|---|---|---|---|
| Plasma 1 | Measured | 1.021 | 0.463 | 0.238 | 0.116 | 0.063 |
| | Expected | | 0.510 | 0.255 | 0.128 | 0.064 |
| | % Recovery | | 91 | 93 | 91 | 99 |
| Plasma 2 | Measured | 7.649 | 3.625 | 2.203 | 1.173 | 0.600 |
| | Expected | | 3.825 | 1.912 | 0.956 | 0.478 |
| | % Recovery | | 95 | 115 | 123 | 125 |
| Plasma 3 | Measured | 1.883 | .990 | 0.471 | 0.228 | 0.105 |
| | Expected | | .942 | 0.471 | 0.235 | 0.118 |
| | % Recovery | | 105 | 100 | 97 | 89 |

| **Adrenaline** ng/mL | Dilution | 1/1 | 1/2 | 1/4 | 1/8 | 1/16 |
|---|---|---|---|---|---|---|
| Sample | | | | | | |
| Urine 1 | Measured | 82.7 | 48.8 | 24.1 | 12.8 | 6.1 |
| | Expected | | 41.4 | 20.7 | 10.3 | 5.2 |
| | % Recovery | | 118 | 116 | 124 | 118 |
| Urine 2 | Measured | 37.0 | 20.4 | 10.5 | 5.1 | 2.8 |
| | Expected | | 18.2 | 9.3 | 4.6 | 2.3 |
| | % Recovery | | 110 | 113 | 111 | 123 |
| Urine 3 | Measured | 25.4 | 12.4 | 6.1 | 3.3 | 1.9 |
| | Expected | | 12.7 | 6.4 | 3.2 | 1.6 |
| | % Recovery | | 98 | 96 | 104 | 121 |
| Plasma 1 | Measured | 0.391 | 0.210 | 0.109 | 0.057 | 0.027 |
| | Expected | | 0.196 | 0.098 | 0.049 | 0.024 |
| | % Recovery | | 107 | 112 | 117 | 110 |
| Plasma 2 | Measured | 0.692 | 0.300 | 0.162 | 0.089 | 0.049 |
| | Expected | | 0.346 | 0.173 | 0.087 | 0.043 |
| | % Recovery | | 87 | 94 | 103 | 113 |
| Plasma 3 | Measured | 2.045 | 0.969 | 0.538 | 0.227 | 0.133 |
| | Expected | | 1.023 | 0.511 | 0.256 | 0.128 |
| | % Recovery | | 95 | 105 | 89 | 104 |

| **Dopamine** ng/mL | Dilution | 1/1 | 1/2 | 1/4 | 1/8 | 1/16 |
|---|---|---|---|---|---|---|
| Sample | | | | | | |
| Urine 1 | Measured | 634.3 | 352.8 | 184.4 | 99.8 | 44.9 |
| | Expected | | 317.2 | 158.6 | 79.3 | 39.6 |
| | % Recovery | | 111 | 116 | 125 | 113 |

| *Dopamine ng/mL* | *Dilution* | *1/1* | *1/2* | *1/4* | *1/8* | *1/16* |
|---|---|---|---|---|---|---|
| Urine 2 | Measured | 157.4 | 65.8 | 39.8 | 18.5 | |
| | Expected | | 78.7 | 39.4 | 19.7 | |
| | % Recovery | | 84 | 101 | 94 | |
| Urine 3 | Measured | 349.4 | 193.2 | 84.5 | 45.4 | 18.2 |
| | Expected | | 174.7 | 87.4 | 43.7 | 21.8 |
| | % Recovery | | 111 | 97 | 104 | 84 |
| Plasma 1 | Measured | 1.324 | 0.554 | 0.314 | 0.167 | 0.099 |
| | Expected | | 0.662 | 0.331 | 0.166 | 0.083 |
| | % Recovery | | 84 | 95 | 101 | 120 |
| Plasma 2 | Measured | 2.011 | 1.011 | 0.545 | 0.252 | 0.135 |
| | Expected | | 1.006 | 0.503 | 0.251 | 0.126 |
| | % Recovery | | 101 | 108 | 100 | 107 |
| Plasma 3 | Measured | 6.830 | 3.171 | 1.968 | 1.047 | 0.458 |
| | Expected | | 3.405 | 1.708 | 0.854 | 0.427 |
| | % Recovery | | 93 | 115 | 123 | 107 |

Reportable range:

| | Urine | Plasma |
|---|---|---|
| Noradrenaline | .960 - 120 ng/mL | 0.050 – 10 ng/mL |
| Adrenaline | 0.320 – 40 ng/mL | 0.030 – 3 ng/mL |
| Dopamine | 7.44 – 1200 ng/mL | 0.060 – 16.7 ng/ml |

Samples may be diluted up to 1:16 if sample concentrations are above the reportable range.

c. *Traceability, Stability, Expected values (controls, calibrators, or methods):*

The calibrators and controls are manufactured using pure substances. The substance is added by weight and the values are confirmed by HPLC.

Stability:

Stability of the kit is limited to the radioactive half-life of the Iodine isotope which is 60 days. Kits with three different lot numbers were stored at 2-8ºC and tested with urine and plasma control material. Testing was performed on the first day, 2-4 weeks and at 6-8 weeks. The results of the mid- an end-testing were compared to the first day testing. The kit demonstrated acceptable stability for 8 weeks.

d. *Detection limit:*

The analytical sensitivity (Limit of Detection, LOD) was determined by calculating 2 SD from the mean of n=20 replicate measurements of the zero standard. The analytical sensitivity was estimated to be 0.008 ng/ml for adrenaline, 0.024 ng/ml for noradrenaline and 0.031 ng/ml for dopamine.

Functional sensitivity was determined from the inter-assay variation coefficient of low samples. The lowest concentration which could be measured with a CV below 20% is Noradrenaline 50 pg/mL, Adrenaline 30 pg/mL and Dopamine 60 pg/mL.

*e. Analytical specificity:*

Cross-reactivity and analytical specificity were assessed by testing compounds whose chemical structure could potentially cause interference. The lists of compounds tested are below:

| | Cross Reactivity (%) | | |
|---|---|---|---|
| Substance | Noradrenaline | Adrenaline | Dopamine |
| Noradrenaline | 100 | 0.6 | 0.013 |
| Adrenaline | <0.03 | 100 | <0.001 |
| Dopamine | 0.09 | <0.001 | 100 |
| DL-Normetanephrine | 0.38 | 0.006 | <0.001 |
| DL-Metanephrine | <0.003 | 0.98 | <0.001 |
| 3-Methoxy-Thyramine | <0.002 | <0.001 | 0.07 |
| DL-Octopamine | <0.005 | <0.001 | <0.001 |
| $OH_3CH_3O$-Phenylpyruvate | <0.002 | <0.001 | <0.001 |
| Adrenochrome | <0.002 | 0.06 | <0.001 |
| DL-Synephrine | <0.001 | 0.15 | <0.001 |

No cross-reactivities were found with Ferule acid, Caffeic acid, Beta-Phenyl Ethylamine, Vanillin, Vanillic mandelic acid, N-Acetyl-L-Tryptophane, Vanillic acid, $OH_3CH3O$-Phenylglycol, Homovanillic-ethanol, Homovanillic acid, 3,4-dihydroxy Mandelic acid and L-Dopa.

Studies were performed to assess common or known substances that could interfere with the method. Highest levels tested with no interference were Hemoglobin 4 mg/dL, Bilirubin 0.4 mg/dL and Triglycerides 22.8 mg/dL.

*f. Assay cut-off:*

Not applicable.

2. Comparison studies:

*a. Method comparison with predicate device:*

Correlation studies were performed comparing the IBL TriCat Radioimmunoassay for Adrenaline, Noradrenaline and Dopamine with the data obtained from HPLC for the same urine and plasma samples. The reference method HPLC is described in Westermann et al. (2002). The correlations were as follows:

| | | | |
|---|---|---|---|
| Adrenaline | Urine | y = 0.99x -3.81 | r = 0.97; n = 60 |
| | Plasma | y = 1.13x -0.05 | r = 0.95; n = 52 |
| Noradrenaline | Urine | y = 0.77x + 16.48 | r = 0.975; n = 57 |
| | Plasma | y = 0.86x -0.02 | r = 0.95; n = 52 |
| Dopamine | Urine | y = 0.96x + 0.71 | r = 0.99; n = 60 |
| | Plasma | y = 0.97x + 0.31 | r = 0.99; n = 33 |

*b. Matrix comparison:*

Not applicable.

3. Clinical studies:

*a. Clinical Sensitivity:*

Not applicable

*b. Clinical specificity:*

Not applicable

c. Other clinical supportive data (when a. and b. are not applicable):

Not applicable

4. Clinical cut-off:

Not applicable

5. Expected values/Reference range:

The following normal range values upper limits have been determined by performing a normal range study. In the study 24h urine samples and plasma samples were collected from normal healthy donors. The limits are in the range of 2 to 3 SD of the mean and as determined by HPLC laboratories. It is recommended that each laboratory establish its own normal range. The following values may serve as a guideline:

| | Urine | | Plasma | |
|---|---|---|---|---|
| | µg/d | nmol/d | pg/mL | nmol/L |
| Adrenaline | < 20 | < 100 | < 100 | < 0.55 |
| Noradrenaline | < 104 | < 615 | < 600 | < 3.55 |
| Dopamine | < 600 | < 3917 | < 100 | < 0.65 |

**N. Proposed Labeling:**

The labeling is sufficient and it satisfies the requirements of 21 CFR Part 809.10.

**O. Conclusion:**

The submitted information in this premarket notification is complete and supports a substantial equivalence decision.